FIG. 1

DNA sequence for human preproparathyroid hormone.

```
        10                  30                  50
ATGATHCCNGCNAARGAYATGGCNAARGTNATGATHGTNATGYTNGCNATHTGYTTYYTN

        70                  90                  110
ACNAARWSNGAYGGNAARWSNGTNAARAARMGNWSNGTNWSNGARATHCARYTNATGCAY

       130                 150                 170
AAYYTNGGNAARCAYYTNAAYWSNATGGARMGNGTNGARTGGYTNMGNAARAARYTNCAR

       190                 210                 230
GAYGTNCAYAAYTTYGTNGCNYTNGGNGCNCCNYTNGCNCCNMGNGAYGCNGGNWSNCAR

       250                 270                 290
MGNCCNMGNAARAARGARGAYAAYGTNYTNGTNGARWSNCAYGARAARWSNYTNGGNGAR

       310                 330
GCNGAYAARGCNGAYGTNAAYGTNYTNACNAARGCNAARWSNCARTRR
```

M = A or C
R = A or G
W = A or T
S = C or G
Y = C or T
H = A or C or T
N = A or G or C or T.

FIG. 2

DNA sequence for human

preproparathyroid hormone in plasmid pSSHPTH-10.

```
         10                                      30                                      50
ATGATGATACCTGCAAAAGACATGGCTAAAGTTATGATTGTCATGTTGGCAATTTGTTTT

         70                                      90                                      110
CTTACAAAATCGGATGGGAAATCTGTTAAGAAGAGATCTGTGAGTGAAATACAGCTTATG

         130                                     150                                     170
CATAACCTGGGAAAACATCTGAACTCGATGGAGAGAGTAGAATGGCTGCGTAAGAAGCTG

         190                                     210                                     230
CAGGATGTGCACAATTTTGTTGCCCTTGGAGCTCCTCTAGCTCCCAGAGATGCTGGTTCC

         250                                     270                                     290
CAGAGGCCCCGAAAAAAGGAAGACAATGTCTTGGTTGAGAGCCATGAAAAAAGTCTTGGA

         310                                     330
GAGGCAGACAAAGCTGATGTGAATGTATTAACTAAAGCTAAATCCCAGTGA
```

FIG. 3

Portion of DNA sequence of the plasmid for insertion into E. coli, coding for human preproparathyroid hormone with flanking sequences.

```
        10                  30                  50
TATGATGATHCCNGCNAARGAYATGGCNAARGTNATGATHGTNATGYTNGCNATHTGYTT

        70                  90                  110
YYTNACNAARWSNGAYGGNAARWSNGTNAARAARMGNWSNGTNWSNGARATHCARYTNAT

       130                 150                 170
GCAYAAYYTNGGNAARCAYYTNAAYWSNATGGARMGNGTNGARTGGYTNMGNAARAARYT

       190                 210                 230
NCARGAYGTNCAYAAYTTYGTNGCNYTNGGNGCNCCNYTNGCNCCNMGNGAYGCNGGNWS

       250                 270                 290
NCARMGNCCNMGNAARAARGARGAYAAYGTNYTNGTNGARWSNCAYGARAARWSNYTNGG

       310                 330                 350
NGARGCNGAYAARGCNGAYGTNAAYGTNYTNACNAARGCNAARWSNCARTRRAAATGAAA

       370                 390                 410
ACAGATATTGTCAGAGTTCTGCTCTAGACAGTGTAGGGCAACAATACATGCTGCTAATTC

       430
AAAGCTCTATTA
```

M = A or C
R = A or G
W = A or T
S = C or T
Y = C or T
H = A or C or T
N = A or G or C or T.

FIG. 4

DNA sequence for human preproparathyroid hormone in plasmid pSSHPTH-10 with flanking sequences.

```
        10                  30                  50
TATGATGATACCTGCAAAAGACATGGCTAAAGTTATGATTGTCATGTTGGCAATTTGTTT

        70                  90                 110
TCTTACAAAATCGGATGGGAAATCTGTTAAGAAGAGATCTGTGAGTGAAATACAGCTTAT

       130                 150                 170
GCATAACCTGGGAAAACATCTGAACTCGATGGAGAGAGTAGAATGGCTGCGTAAGAAGCT

       190                 210                 230
GCAGGATGTGCACAATTTTGTTGCCCTTGGAGCTCCTCTAGCTCCCAGAGATGCTGGTTC

       250                 270                 290
CCAGAGGCCCCGAAAAAAGGAAGACAATGTCTTGGTTGAGAGCCATGAAAAAAGTCTTGG

       310                 330                 350
AGAGGCAGACAAAGCTGATGTGAATGTATTAACTAAAGCTAAATCCCAGTGAAAATGAAA

       370                 390                 410
ACAGATATTGTCAGAGTTCTGCTCTAGACAGTGTAGGGCAACAATACATGCTGCTAATTC

       430
AAAGCTCTATTA.
```

FIG. 5

DNA sequence coding for preproparathyroid hormone in pSSHPTH-10 with flanking sequences, showing the corresponding amino acid sequence of preproparathyroid hormone.

```
        10                  30                  50
TATGATGATACCTGCAAAAGACATGGCTAAAGTTATGATTGTCATGTTGGCAATTTGTTT
    MetIleProAlaLysAspMetAlaLysValMetIleValMetLeuAlaIleCysPh

        70                  90                  110
TCTTACAAAATCGGATGGGAAATCTGTTAAGAAGAGATCTGTGAGTGAAATACAGCTTAT
eLeuThrLysSerAspGlyLysSerValLysLysArgSerValSerGluIleGlnLeuMe

       130                 150                 170
GCATAACCTGGGAAAACATCTGAACTCGATGGAGAGAGTAGAATGGCTGCGTAAGAAGCT
tHisAsnLeuGlyLysHisLeuAsnSerMetGluArgValGluTrpLeuArgLysLysLe

       190                 210                 230
GCAGGATGTGCACAATTTTGTTGCCCTTGGAGCTCCTCTAGCTCCCAGAGATGCTGGTTC
uGlnAspValHisAsnPheValAlaLeuGlyAlaProLeuAlaProArgAspAlaGlySe

       250                 270                 290
CCAGAGGCCCCGAAAAAAGGAAGACAATGTCTTGGTTGAGAGCCATGAAAAAAGTCTTGG
rGlnArgProArgLysLysGluAspAsnValLeuValGluSerHisGluLysSerLeuGl

       310                 330                 350
AGAGGCAGACAAAGCTGATGTGAATGTATTAACTAAAGCTAAATCCCAGTGAAAATGAAA
yGluAlaAspLysAlaAspValAsnValLeuThrLysAlaLysSerGlnEnd

       370                 390                 410
ACAGATATTGTCAGAGTTCTGCTCTAGACAGTGTAGGGCAACAATACATGCTGCTAATTC

       430
AAAGCTCTATTA.
```

Figure 6. Nucleotide sequence of the MF 1-HPTH fusion gene from pS LX5-HPTH1. Nucleotide nos. 1-173 makeup the MF 1 promoter region and 5' noncoding sequence. 174-440 is the MF 1 N-terminal coding sequence. 441-695 is the HPTH sequence obtained from pSSHPTH-10. 696-726 is an HPTH 3' noncoding sequence from pSSHPTH-10. 727-732 is from pUC19. 733-874 is MF 1 3' noncoding sequence and transcriptional termination signal.

```
          10                  30                  50
AGTGCAAGAAAACCAAAAAGCAACAACAGGTTTTGGATAAGTACATATATAAGAGGGCCT

          70                  90                  110
TTTGTTCCCATCAAAAATGTTACTGTTCTTACGATTCATTTACGATTCAAGAATAGTTCA

          130                 150                 170
AACAAGAAGATTACAAACTATCAATTTCATACACAATATAAACGACCAAAAGAATGAGAT

          190                 210                 230
TTCCTTCAATTTTTACTGCAGTTTTATTCGCAGCATCCTCCGCATTAGCTGCTCCAGTCA

          250                 270                 290
ACACTACAACAGAAGATGAAACGGCACAAATTCCGGCTGAAGCTGTCATCGGTTACTCAG

          310                 330                 350
ATTTAGAAGGGGATTTCGATGTTGCTGTTTTGCCATTTTCCAACAGCACAAATAACGGGT

          370                 390                 410
TATTGTTTATAAATACTACTATTGCCAGCATTGCTGCTAAAGAAGAAGGGGTATCTTTGG

          430                 450                 470
ATAAAAGAGAGGCTGAAGCTTCTGTGAGTGAAATACAGCTTATGCATAACCTGGGAAAAC

          490                 510                 530
ATCTGAACTCGATGGAGAGAGTAGAATGGCTGCGTAAGAAGCTGCAGGATGTGCACAATT

          550                 570                 590
TTGTTGCCCTTGGAGCTCCTCTAGCTCCCAGAGATGCTGGTTCCCAGAGGCCCCGAAAAA

          610                 630                 650
AGGAAGACAATGTCTTGGTTGAGAGCCATGAAAAAAGTCTTGGAGAGGCAGACAAAGCTG

          670                 690                 710
ATGTGAATGTATTAACTAAAGCTAAATCCCAGTGAAAATGAAAACAGATATTGTCAGAGT
```

730 750 770

TCT TCTAGAGTCGACTTTGTTCCCACTG CTTTTAGCTCGTACAAAATACAATATAC

790 810 830

TTTTCATTTCTCCGTAAACAACCTGTTTTCCCATGTAATATCCTTTTCTATTTTTCGTTT

10 850 870

CGTTACCAACTTTACACATACTTTATATAGCTAT

Figure 7. Partial DNA sequence for the plasmid for insertion into yeast in which: Nucleotide nos. 1-173 mak up the MF 1 promoter region and 5' noncoding sequence. 174-440 is the MF 1 N-terminal coding sequence. 441-695 is an HPTH sequence. 696-726 is an HPTH 3' noncoding sequence from pSSHPTH-10. 727-732 is from pUC19. 733-874 is MF 1 3' noncoding sequence and transcriptional termination signal.

```
10                  10                                  30
50
AGTGCAAGAAAACCAAAAAGCAACAACAGGTTTTGGATAAGTACATATATAAGAGGGCCT

                    70                  90                  110
TTTGTTCCCATCAAAAATGTTACTGTTCTTACGATTCATTTACGATTCAAGAATAGTTCA

                    130                 150                 170
AACAAGAAGATTACAAACTATCAATTTCATACACAATATAAACGACCAAAAGAATGAGAT

                    190                 210                 230
TTCCTTCAATTTTTACTGCAGTTTTATTCGCAGCATCCTCCGCATTAGCTGCTCCAGTCA

                    250                 270                 290
ACACTACAACAGAAGATGAAACGGCACAAATTCCGGCTGAAGCTGTCATCGGTTACTCAG

                    310                 330                 350
ATTTAGAAGGGGATTTCGATGTTGCTGTTTTGCCATTTTCCAACAGCACAAATAACGGGT

                    370                 390                 410
TATTGTTTATAAATACTACTATTGCCAGCATTGCTGCTAAAGAAGAAGGGGTATCTTTGG

                    430                 450                 470
ATAAAAGAGAGGCTGAAGCTWSNGTNWSNGARATHCARYTNATGCAYAAYYTNGGNAARC

                    490                 510                 530
AYYTNAAYWSNATGGARMGNGTNGARTGGYTNMGNAARAARYTNCARGAYGTNCAYAAYT

                    550                 570                 590
TYGTNGCNYTNGGNGCNCCNYTNGCNCCNMGNGAYGCNGGNWSNCARMGNCCNMGNAARA

                    610                 630                 650
ARGARGAYAAYGTNYTNGTNGARWSNCAYGARAARWSNYTNGGNGARGCNGAYAARGCNG

                    670                 690                 710
AYGTNAAYGTNYTNACNAARGCNAARWSNCARTRRAAATGAAAACAGATATTGTCAGAGT
```

730 750 770
TCT(:CTAGAGTCGACTTTGTTCCCACTGT. TTTTAGCTCGTACAAAATACAATATAC

790 810 830
TTTTCATTTCTCCGTAAACAACCTGTTTTCCCATGTAATATCCTTTTCTATTTTTCGTTT

850 870
CGTTACCAACTTTACACATACTTTATATAGCTAT, wherein

M = A or C
R = A or G
W = A or T
S = C or G
Y = C or T
H = A or C or T
N = A or G or C or T

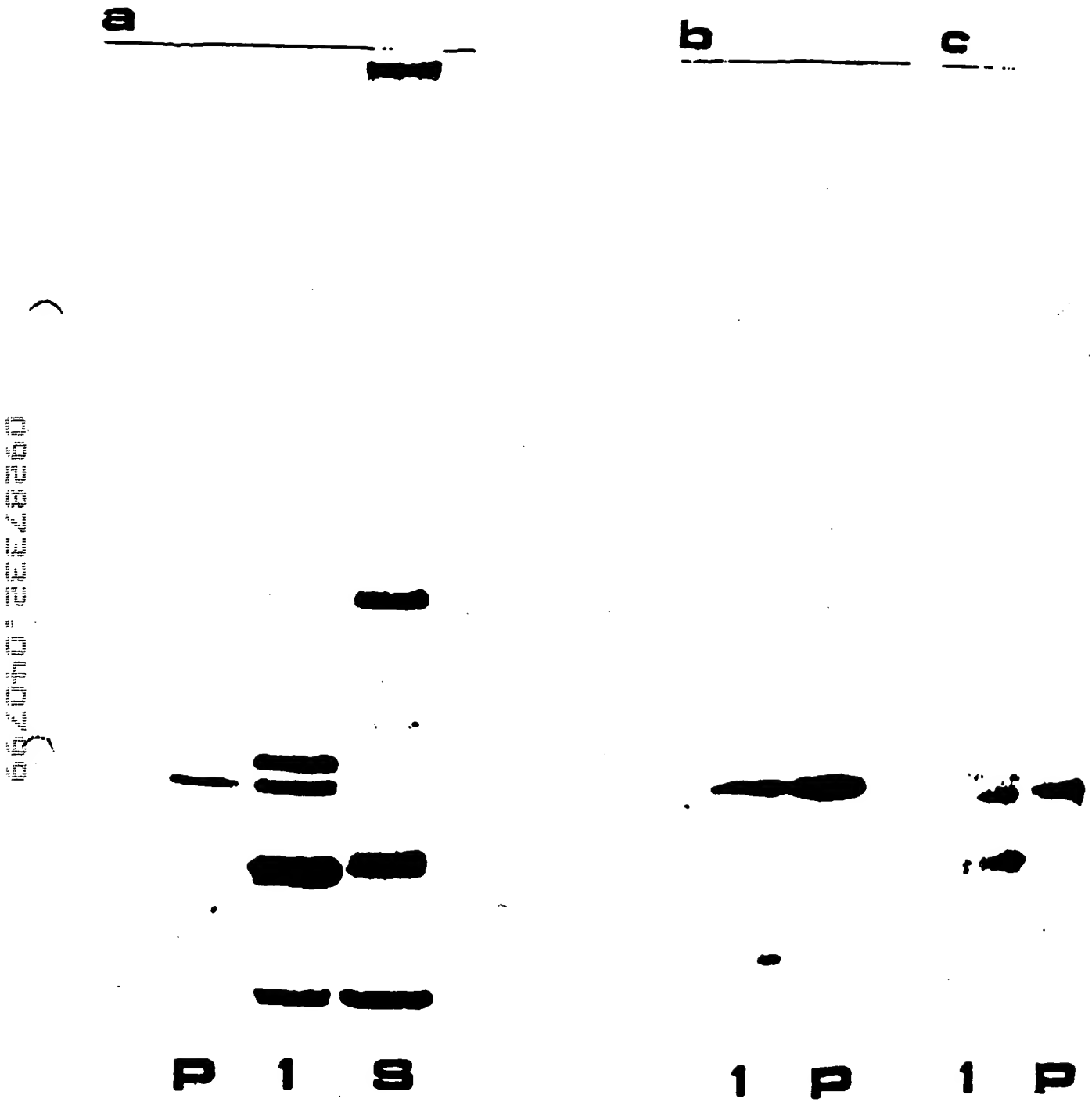
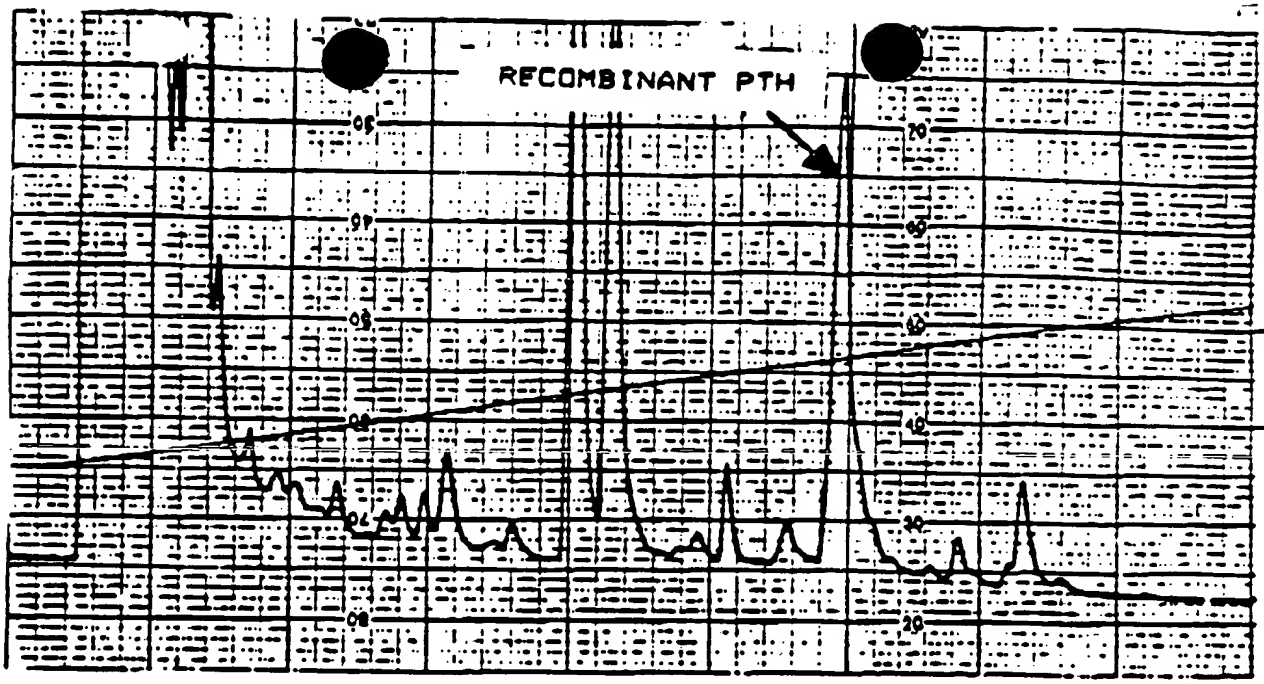


FIG. 8


RECOMBINANT PTH

B.

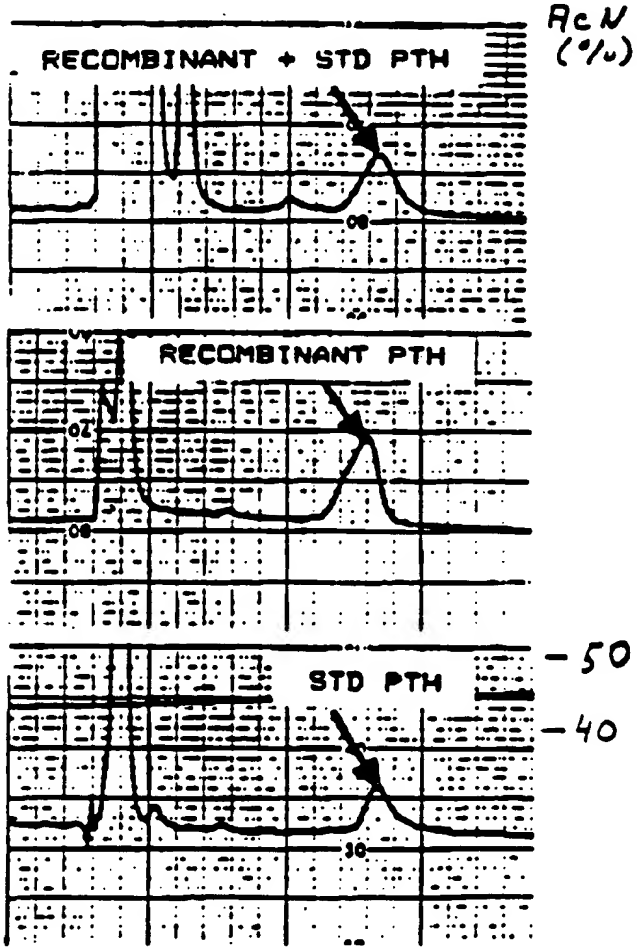

RECOMBINANT + STD PTH
RECOMBINANT PTH
STD PTH

FIG. 9

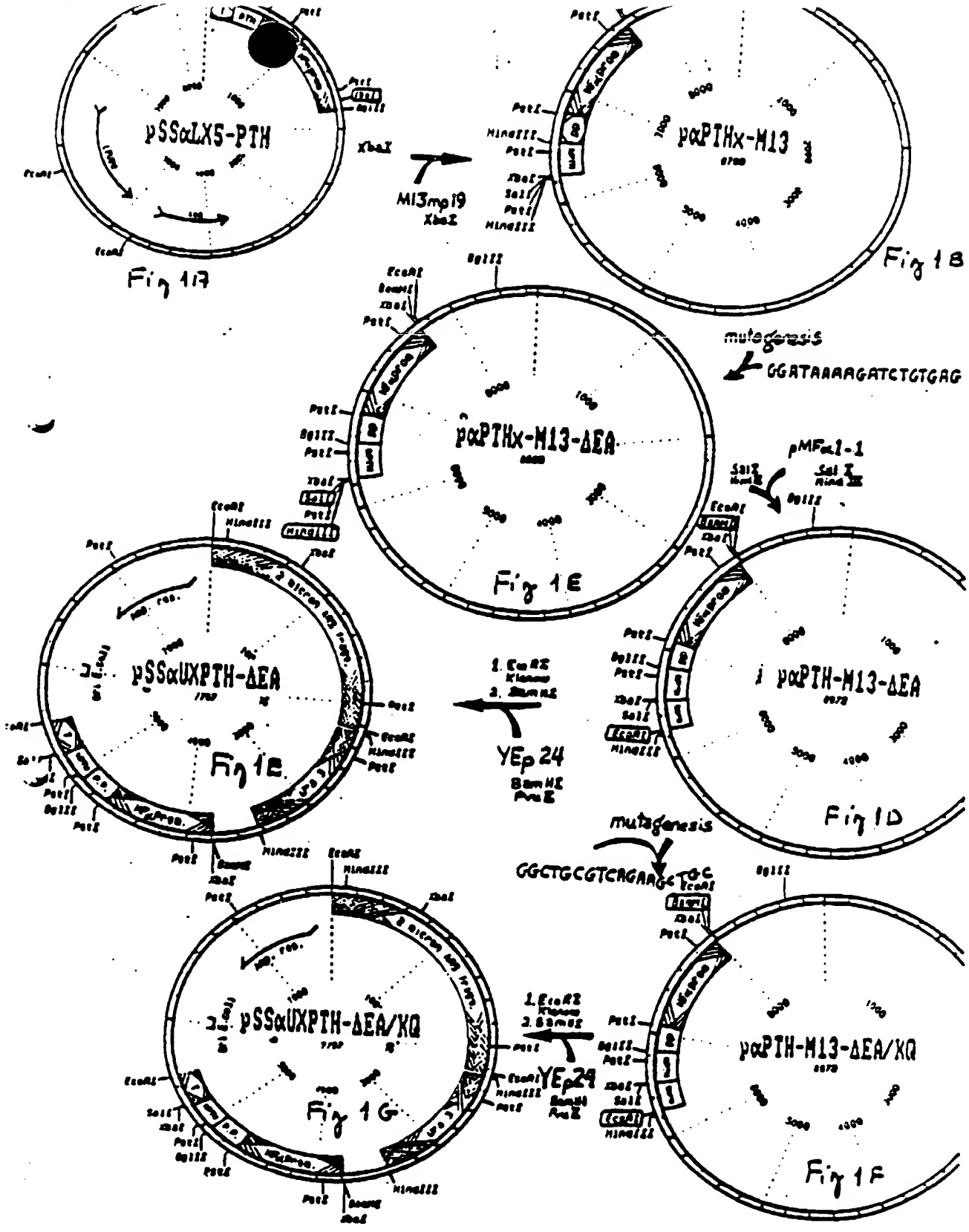

pSSαLX5-PTH
Fig 1A
M13mp19
XbaI
pαPTHx-M13
Fig 1B
mutagenesis
GGATAAAAGATCTGTGAG
pαPTHx-M13-ΔEA
Fig 1E
pMFα1-1
pSSαUXPTH-ΔEA
Fig 1C
YEp 24
pαPTH-M13-ΔEA
Fig 1D
mutagenesis
GGCTGCGTCAGAAGCTGC
pSSαUXPTH-ΔEA/KQ
Fig 1G
YEp24
pαPTH-M13-ΔEA/KQ
Fig 1F

FIG. 10

In vitro mutagenesis of hPTH

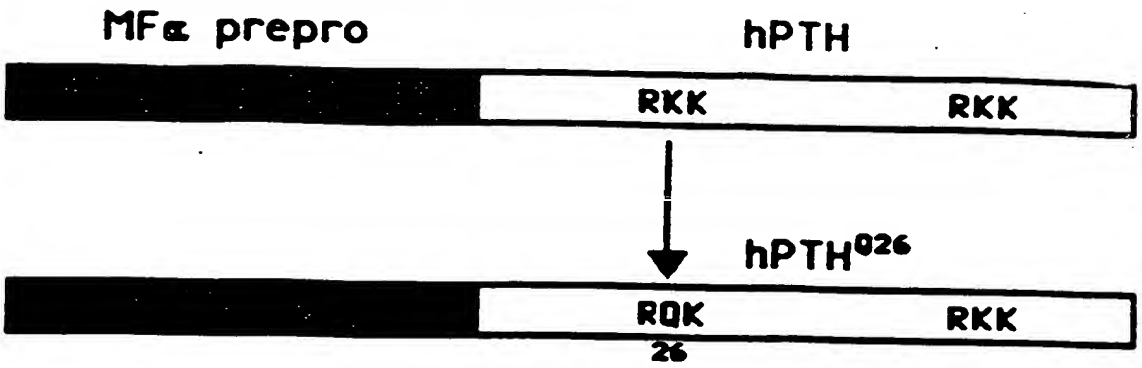
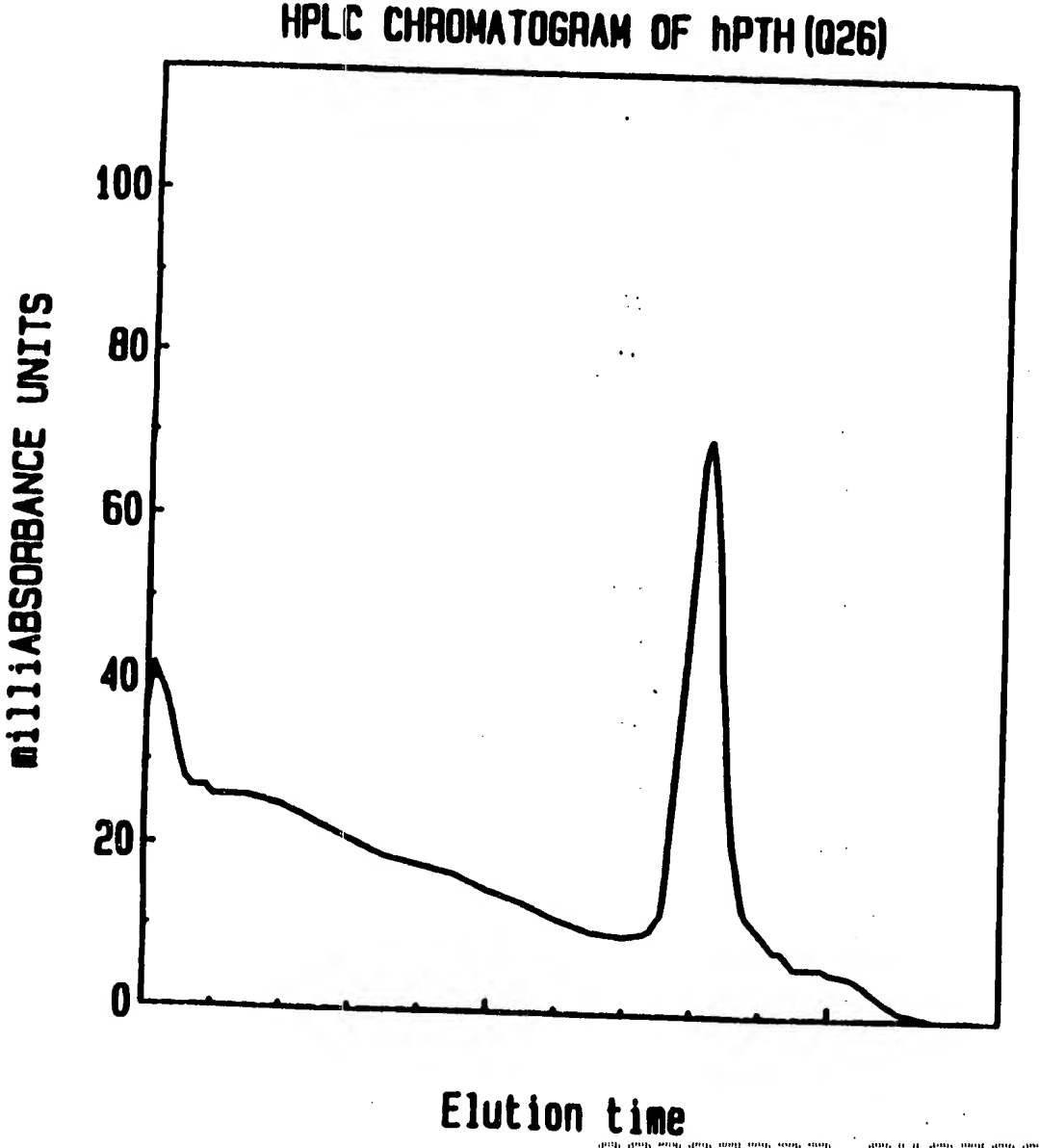


FIG. 11

FIG. 12

A



B

FIG. 13

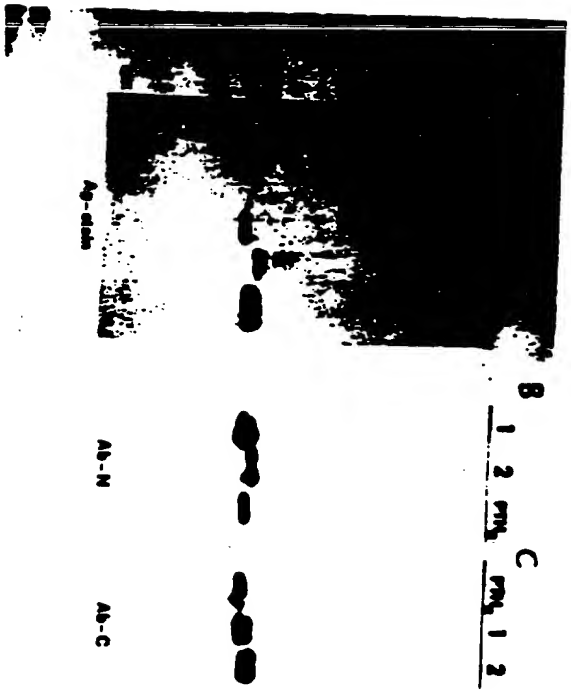
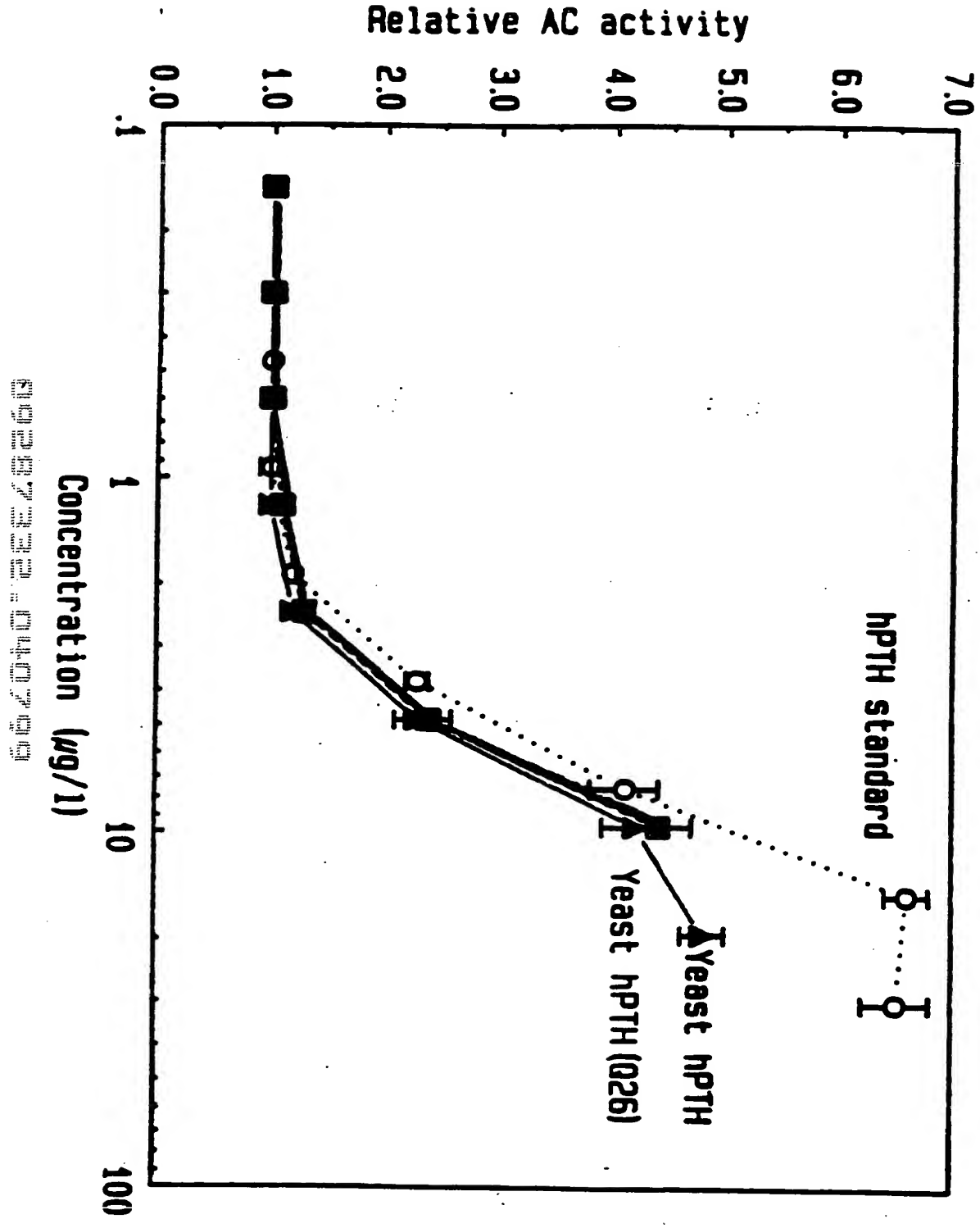


FIG. 14

FIG. 15


Relative AC activity
0.0
1.0
2.0
3.0
4.0
5.0
6.0
7.0
.1
1
10
100
Concentration (µg/l)
hPTH standard
Yeast hPTH
Yeast hPTH (026)